

## YIELD OF FOUR *Agaricus bisporus* STRAINS IN THREE COMPOST FORMULATIONS AND CHEMICAL COMPOSITION ANALYSES OF THE MUSHROOMS

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### ABSTRACT

Three compost formulations, consisting of two varieties of *Cynodom dactylon* (L.) Pers. (Coast-cross and Tyfton) and oat (*Avena sativa*) straw were tested for the cultivation of *A. bisporus* strains ABI-01/01, ABI-04/02, ABI-05/03, and ABI-06/04. A completely randomized experimental design in a factorial scheme was adopted, with 12 treatments (4 *A. bisporus* strains × 3 types of compost) and 8 replicates. Each experimental unit corresponded to one box containing 12 – 12.5 kg fresh wet compost. The data were submitted to analysis of variance and the means were compared by Tukey test. According to the results, productivity of mushrooms was influenced by strain and/or compost type. It was also verified that crude protein, ash, and crude fiber contents in the mushroom varied with *A. bisporus* strain and straw used in the formulation of the compost.

**Key-words:** *Agaricus bisporus*, strains, straw, nutritional composition, productivity.

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### INTRODUCTION

The formulation of compost is crucial to carry on composting for *Agaricus* sp and it is essential to an adequate balance between nutrients, especially carbon and nitrogen (21). As a result, mycelium growth can be influenced by a reduction or excess concentration of nutrients, accumulation of toxic products, production of secondary metabolites, and changes in factors such as pH, which consequently can influence the mushroom production (2,14,15).

Several formulations can be used for production of *Agaricus* sp. which are usually determined by the availability and cost of substrates (16). Straws (rice, wheat, oat and barley), by-products (sugarcane bagasse), horse and chicken manure are the most important components used as lignocellulosic sources in composts (11,12). However, due to a limited availability of horse manure in large amounts around the world, composts referred to as “synthetic” have been increasingly used, whose formulations are prepared without the addition of it (7).

Straw of cereals are used in most countries as a basic material for composts and wheat straw is commonly used in Europe and the United States (5) while rice straw is largely used in Asian countries (6).

In Brazil, wheat, rice, brachiaria, coast-cross, tyfton, barley straw have been used as substrates in formulations for composts of *Agaricus* sp (11). In some regions of São Paulo State, suppliers of straw (e.g. coast-cross, tyfton and *Brachiaria* sp.) provide in a very accessible price that are not sold because of their low quality standards for animal feed but still in a good quality for composting of mushroom cultivation. Usually supplying substrates for mushroom composts vary according to availability during the seasons of the year.

However, few scientific studies have compared the effectiveness of these substrates on the productivity of *A. bisporus* strains, where inconsistent information about advantages in using specific straws for mushroom compost are provide through mushroom farmers.

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Many edible mushrooms constitute a high-quality food, and are rich in proteins, chitin, vitamins, and minerals, and poor in calories and fat (9, 13, and 20). However, the nutritional composition of mushrooms varies depending on the species, strain, processing and development stage of the mushrooms (9,10).

Thus, this study aimed to characterize the nutritional composition of mushroom from *A. bisporus* strains cultivated in composts prepared with three different straws as well as to evaluate the productivity and biological efficiency.

## MATERIAL AND METHODS

The experiment was carried out in the facilities of Módulo de Cogumelos (Mushroom Research Center), Faculdade de Ciências Agrônomicas (College of Agronomic Sciences), UNESP (São Paulo State University), Botucatu/ SP, Brazil.

### Spawn production

The study was carried out with four strains from commercial cultivations maintained in the Culture Collection of the Mushroom Center of FCA/UNESP. Strains ABI-01/01 (medium size, white, steam thin and long, precocious opening cap, isolated in Mogi das Cruzes/SP), ABI-04/02 (medium size, white, high productivity, provided by Brasmicel-Mogi das Cruzes/ SP; ABI-05/03 (medium-big size, white, isolated from Cabriúva/ SP); and ABI-06/04 (medium size, white, summer strain, producing up 20-22°C, isolated from Itú/ SP). They were stocked in tubes with CA (compost-agar) medium submerged in sterilized mineral oil and preserved at 8°C.

Initially, disks of the mother spawn were transferred to Petri dishes with CA medium under aseptic conditions; after inoculation, the Petri dishes were transferred to an incubator and kept for 25 days at 22±1°C for colonization. They were transferred to flasks with sorghum grains (400g), gypsum, and calcium carbonate. The grains were initially boiled in water for 40 minutes. After draining the excess water, 20g kg<sup>-1</sup> calcium carbonate and 160g kg<sup>-1</sup> gypsum were added (d.w). The bottom part of the flask lid was fitted with a paper filter. The bags were incubated at 22 ± 1°C for 20 days. The spawn was produced by placing the substrate in high-density polyethylene bags (HDPE), using about 1200g of grains per bag. The bags contained Tyvek® tag in their upper part, thus allowing gas exchanges. In both cases, the substrates were autoclaved at 121°C for 3 hours. After incubation at 22 ± 1°C for 30 days, the substrates were already colonized by the fungus, being thus referred to as spawn, and were then ready to be inoculated into the compost. The substrates used for the composts were analyzed at FCA/UNESP Fertilizers Laboratory and three types of compost were formulated, consisting of coast-cross, tyfton or oat straw in order to obtain a initial C/N ratio of 25/1 (Table 1).

**Table 1.** Formulation of composts prepared with coast-cross, tyfton, or oat straw.

Ingredients, kg	Compost					
	Coast-cross		Tyfton		Oat	
	FW	DW	FW	DW	FW	DW
Sugarcane bagasse	600.0	300.0	600.0	300.0	600.0	300.0
Coast-cross straw	280.0	252.0				
Tifton straw			280.0	243.6		
Oat straw					280.0	243.6
Soybean meal	100.0	90.0	90.0	81.0	100.0	90.0
Urea	4.0	4.0	4.0	4.0	4.0	4.0
Weight of compost, kg	984.0	646.0	974.0	628.6	984.0	637.6
Initial carbon, kg		318.1		318.1		321.4
Initial nitrogen, kg		12.5		12.8		12.6
Initial C/N ratio		25.4		24.9		25.6

\*FW= fresh weight and DW= dry weight.

Composting phase I was performed on concrete floor, open sides and natural ventilation. Before forming the windrows, all straw types were moistened and turned every day for a total period of 8 days. The windrows were formed piling a straw layer (20 cm in height), followed by a sugarcane bagasse layer (20 cm in height) and built into a 1.8 m height × 1.8 m width. Urea and soybean meal were added equitably to the windrow. The composts were turned each other day, and water was added manually, with a hose, to maintain the moisture in 70-75%. In all, 5 turnings were performed, totaling 12 days in Phase I. In phase II, the composts were transferred to perforated plastic boxes, measuring 56.5 × 46.5 × 28.5 cm (length, width, and height). The boxes were randomly arranged inside a chamber (Dalsem mushrooms) for pasteurization (6 hours at 61±1°C) and conditioning (11 days at 48±1°C). At the end of phases I and II, three samples of each compost were collected and dehydrated at 65°C for 48 hours for analyses of carbon and nitrogen and pH. Results are shown in Table 2.

### Inoculation and spawning run

Inoculation of the composts were made manually, by adding 1.5g of *A. bisporus* spawn per kilogram f.w. compost. To accomplish this, the composts were apportioned and transferred to other smaller polyethylene boxes (12 – 12.5 g of f.w. compost) internally lined with clear polyethylene plastic film containing holes on the bottom. The boxes were randomly arranged in an incubator (Dalsem Mushroom) and kept for 13 days at 22±1°C. A loamy oxisol-alfisol from Fazenda Lageado (FCA/UNESP) was used as casing soil. The soil pH was corrected to 7.0 by adding calcium carbonate (30 kg m<sup>-3</sup>) 20 days before casing. The soil was moistened to 60%. 30% wood charcoal (1 to 2 cm thick)

**Table 2.** Moisture, carbon, and nitrogen contents, C/N ratio, and pH of composts prepared with coast-cross, tyfton, or oat at the end of composting phases I and II.

	Compost		
	Coast-cross	Tifton	Oat
	End of phase I		
M*	71.10	71.13	74.74
C	44.33	44.67	44.67
N	2.48	2.05	2.18
C/N	17.85	21.82	20.46
pH	6.27	6.00	6.13
	End of phase II		
M*	69.20	71.52	70.70
C	40.33	40.33	43.33
N	2.24	2.17	2.44
C/N	17.98	18.59	17.74
pH	6.60	6.83	6.60

\*M = moisture; C = carbon; N = nitrogen.

were added and pasteurization was carried out at 62°C for 8 hours, in a Dalsem Mushrooms container. Each box received about 15 kg of casing soil (4 cm height) and watered to keep in 70% of moisture. The compost was covered with a transparent plastic, placed in darkness and incubated for 15 days at 22±1°C. After the casing soil was colonized, the plastics were removed and maintained temperature at 22±1°C and relative humidity between 75 and 85%.

#### Variables analyzed

**Number and fresh weight of mushroom:** The number (NM) and fresh weight of mushroom (FWM) were determined daily during the harvesting.

**Productivity and biological efficiency:** Productivity (P) was expressed as fresh weight of mushrooms (FWM)/fresh weight of compost (FWC) x 100 and biological efficiency (BE%) as the fresh weight of mushrooms (FWM)/dry weight of compost (DWC) x 100. The FWM was determined at the end of the harvesting and FWC was determined at the end of phase II.

**Nutritional analysis of mushroom from *A. bisporus* strains:** The nutritional analyses of the mushroom were performed at Faculdade de Medicina Veterinária e Zootecnia - FMVZ/UNESP's Bromatology Laboratory, Botucatu-SP. Two samples of dehydrated mushroom were collected at the 1<sup>st</sup> flush and were determined crude protein, fat, ash, and crude fiber contents, according to Silva and Queiroz (18).

#### Experimental Design and Statistical Analysis

A completely randomized experimental design in a factorial scheme was adopted, with 12 treatments (4 *A. bisporus* strains × 3 types of compost) and 8 replicates. The data were submitted to analysis of variance and the means were compared by Tukey test (19), using the Sisvar 4.2 statistical program, developed by Department of Mathematical Sciences from UFPA (Federal University of Lavras), MG.

#### RESULTS AND DISCUSSION

According to Table 3, there was an effect of strain, straw and of the strain × straw interaction for the biological efficiency. However, for productivity and fresh weight of mushroom there was an effect of straw and of the strain × straw interaction. Only a straw effect was observed for number of mushroom.

The values for number and weight of mushroom, productivity, and biological efficiency of *A. bisporus* strains ABI-05/03, ABI-06/04, ABI-04/02, and ABI-01/01 are shown in Table 4. The number of mushroom in composts derived from coast-cross and oat straw did not vary with *A. bisporus* strain. For the tyfton compost, however, strain ABI-04/02 was poorer than strain ABI-01/01. With regard to number of mushroom, there was no compost effect in strains ABI-06/04 and ABI-01/01. On the other hand, strain ABI-05/03 produced a smaller number of mushrooms when grown in tyfton compost as compared to the oat compost. A similar fact occurred with strain ABI-04/02, which produced significantly poorer results when cultivated in tyfton compost as compared with the oat and coast-cross composts.

With regard to fresh weight of mushrooms according to strains, there was no significant difference between strains when inoculated in coast-cross and oat composts (Table 4). On the other hand, when grown in the tyfton compost, strain ABI-04/02 was significantly poorer than strain ABI-01/01.

**Table 3.** F values obtained in the analysis of variance for number of mushroom (NM), fresh weight of mushroom (FWM), productivity (P), and biological efficiency (BE) of *Agaricus bisporus* strains ABI-05/03, ABI-06/04, ABI-04/02, and ABI-01/01, cultivated in three different composts prepared with coast-cross, tyfton, or oat straw.

Causes of variation	NM	FWM	P	BE
Strain (St)	2.19 ns	2.60 ns	2.66 ns	2.73*
Straw (S)	7.79**	6.76**	6.96**	5.05**
St x S	1.62 ns	2.33*	2.33*	2.40*
CV %	35.48	33.05	32.87	32.97

\*\*significance <1% level, \*significance <5% level, ns: not significantly different. CV%: coefficient of variation.

**Table 4.** Total mean values for number and fresh weight of mushroom, productivity, and biological efficiency of *Agaricus bisporus* strains ABI-05/03, ABI-06/04, ABI-04/02, and ABI-01/01 depending on the straw type used in the compost.

Strains	Straw		
	Coast-cross	Tyfton	Oat
Number of mushrooms			
ABI-05/03	183 A ab*	140 AB b	232 A a
ABI-06/04	236 A a	161 AB a	194 A a
ABI-04/02	190 A a	96 B b	208 A a
ABI-01/01	203 A a	210 A a	225 A a
Fresh weight of mushrooms (g)			
ABI-05/03	1723.8 A ab	1219.3 AB b	1898.3 A a
ABI-06/04	2205.3 A a	1572.1 AB a	1680.8 A a
ABI-04/02	1637.4 A a	851.9 B b	1829.9 A a
ABI-01/01	1662.3 A a	1871.4 A a	1906.3 A a
Productivity (%)			
ABI-05/03	14.0 A ab	9.8 AB b	15.4 A a
ABI-06/04	18.0 A a	12.8 AB a	13.8 A a
ABI-04/02	13.3 A a	7.0 B b	15.0 A a
ABI-01/01	13.6 A a	15.3 A a	15.5 A a
Biological efficiency (%)			
ABI-05/03	45.3 A a	34.5 AB a	52.5 A a
ABI-06/04	58.3 A a	44.9 A a	47.2 A a
ABI-04/02	43.3 A a	24.4 B b	51.2 A a
ABI-01/01	44.1 A a	53.4 A a	52.9 A a

\*Means followed by the same upper case letters within a column and lower case letters within a row are not significantly different (Tukey, 5%).

There was no compost type effect on fresh weight of mushroom for strains ABI-06/04 and ABI-01/01. Strain ABI-05/03, however, had a poorer performance when cultivated in the tyfton compost when compared with the oat compost. Finally, strain ABI-04/02 produced a significantly lower result when grown in the tyfton compost as compared with the results obtained in the oat and coast-cross straw composts. Dias *et al.* (3) analyzed the production of *Pleurotus sajor-caju* in different agricultural residues (bean straw corn straw and coffee hulls) and also observed mycelium growth, production, and biological efficiency differences depending on the compost used.

Comparing the productivity of the strains on composts, it was observed that significant differences existed between strains in the coast-cross and oat composts. In the tyfton compost, strain ABI-04/02 was poorer than strain ABI-01/01. There was no compost type effect on productivity for strains ABI-06/04

and ABI-01/01 (Table 4). On the other hand, strain ABI-05/03 produced a significantly lower result when grown in the tyfton compost as compared with the results obtained in the oat compost. A similar fact occurred with strain ABI-04/02, which gave significantly lower results when inoculated in the tyfton compost as compared with the oat and coast-cross composts.

The biological efficiency of *A. bisporus* strains in the coast-cross and oat composts could not be discriminated. Strain ABI-04/02 inoculated in the tyfton compost, however, had a significantly lower value in relation to strains ABI-06/04 and ABI-01/01 (Table 4). In comparing the performance of each strain in the different types of composts tested, it was observed that the performances of strains ABI-05/03, ABI-06/04, and ABI-01/01 were the same in the three types of composts. Nevertheless, strain ABI-04/02 had a significantly poorer result when cultivated in the tyfton compost.

Table 5 presents the F values obtained in the analysis of variance for crude protein, fat, ash, and crude fiber of *Agaricus bisporus* strains ABI-05/03, ABI-06/04, ABI-04/02, and ABI-01/01 cultivated in the three types of composts. There were effects of strain, straw and of the line × straw interaction on variables crude protein, ash, and crude fiber. However, there was no effect of the factors and of the interaction between them on fat.

The crude protein, fat, ash, and crude fiber values for *Agaricus bisporus* strains ABI-05/03, ABI-06/04, ABI-04/02, and ABI-01/01 are displayed in Table 6. In regard to crude protein content, all *A. bisporus* strains in the coast-cross straw compost were different among themselves. The best performance was achieved by strain ABI-04/02 and the poorest by strain ABI-01/01. In the tyfton straw compost, however, crude protein content was inferior in strain ABI-01/01 in relation to the other strains, which were similar among themselves. Finally, when grown in the oat compost, the best performance was observed for strain ABI-04/02 and the poorest for strain ABI-01/01. On the other hand, in comparing crude protein content for the various

**Table 5.** F values obtained in the analysis of variance for crude protein, fat, ash, and crude fiber of *Agaricus bisporus* strains ABI-05/03, ABI-06/04, ABI-04/02, and ABI-01/01, cultivated in three types of composts prepared from coast-cross, tyfton, or oat straw.

Causes of variation	Crude Protein	Fat	Ash	Crude fiber
Strain (St)	173.85**	0.54 ns	44.48 **	43.17**
Straw (T)	14.03**	3.73 ns	11.21**	21.75**
S x T	19.59**	0.60 ns	4.41*	8.60**
CV%	1.65	18.00	1.69	4.92

\*\*significance <1% level; \*significance <5% level; ns: not significantly different; CV%: coefficient of variation.

compost types, strains ABI-05/03 and ABI-04/02 gave the best performances when grown in the coast-cross compost. Strains ABI-06/04 and ABI-01/01, however, showed the best results when cultivated in the tyfton compost (Table 6).

As to fat, which varied from 1.90 to 3.06%, there was no difference due to strains or straw types.

With regard to ash content, strain ABI-01/01 had the poorest performance in relation to the others when cultivated in the coast-cross and oat composts. A similar result also occurred when this strain was cultivated in the tyfton compost, with the poorest performance when compared with strains ABI-06/04 and ABI-04/02. There was no compost type effect on ash content for strain ABI-05/03. Strains ABI-06/04 and ABI-01/01, however, had the poorest result when grown in the oat compost, as compared with the tyfton compost. Finally, strain ABI-04/02 obtained the poorest performance when cultivated in the coast-cross compost in relation to the tyfton compost.

**Table 6.** Mean crude protein, fat, ash, and crude fiber content values for *Agaricus bisporus* strains ABI-05/03, ABI-06/04, ABI-04/02, and ABI-01/01 in different compost types.

Strains	Type of compost		
	Coast-cross	Tifton	Oat
Crude Protein (%)			
ABI-05/03	36.17 B a*	34.36 A b	34.30 B b
ABI-06/04	32.86 C b	35.21 A a	33.69 B b
ABI-04/02	37.88 A a	34.30 A c	36.28 A b
ABI-01/01	29.80 D b	31.37 B a	26.78 C c
Fat (%)			
ABI-05/03	2.33 A a	2.05 A a	2.34 A a
ABI-06/04	2.36 A a	2.34 A a	2.64 A a
ABI-04/02	2.36 A a	2.17 A a	3.06 A a
ABI-01/01	1.90 A a	2.23 A a	2.86 A a
Ash (%)			
ABI-05/03	11.39 A a	11.09 BC a	11.14 AB a
ABI-06/04	11.12 A ab	11.33 AB a	10.71 B b
ABI-04/02	11.02 A b	11.77 A a	11.36 A ab
ABI-01/01	10.31 B ab	10.60 C a	9.83 C b
Crude Fiber (%)			
ABI-05/03	7.56 A b	7.33 A b	10.31 A a
ABI-06/04	6.86 A a	7.01 AB a	7.42 B a
ABI-04/02	7.50 A a	7.60 A a	7.56 B a
ABI-01/01	5.64 B a	6.07 B a	6.57 B a

\*For each variable, means followed by the same upper case letters in each column and lower case letters in each row do not differ (Tukey, 5%).

In comparing the crude fiber content among the strains, strain ABI-01/01, cultivated in the coast-cross compost, gave the poorest result in relation to the other strains. A similar fact occurred in the tyfton compost, where this strain had an inferior performance than strains ABI-05/03 and ABI-04/02. Finally, in the oat compost, strain ABI-05/03 had a superior result in relation to the other strains. On the other hand, in comparing crude fiber content for the various compost types, it was observed that strain ABI-05/03 showed the best performance when cultivated in the oat compost. For strains ABI-06/04, ABI-04/02, and ABI-01/01, however, crude fiber contents did not vary with compost (Table 6).

Maio *et al.* (8) studied the influence of substrate composition on the growth velocity of the mushroom *Pleurotus ostreatus* and observed a significant correlation between substrate formulation and the nutritional composition of the mushroom. Shibata and Demiate (17) evaluated the chemical composition of the mushroom *Agaricus blazei* and concluded that there are nutritional differences between the two *A. blazei* strains tested, with strain AbM being richer in fibers and minerals, while strain Jun-17 showed a higher carbohydrate content.

## RESUMO

### Produção de quatro linhagens de *Agaricus bisporus* em três formulações de compostos e análises bromatológicas dos cogumelos produzidos

Três formulações de composto, à base de palhas de *Cynodom dactylon* (L.) Pers. (cultivares Coast-cross e Tyfton) e Aveia-*Avena sativa*, foram testadas no cultivo das linhagens ABI-01/01, ABI-04/02, ABI-05/03 e ABI-06/04 de *A. bisporus*. O delineamento experimental foi em esquema fatorial, inteiramente casualizado com 12 tratamentos (4 linhagens de *A. bisporus* x 3 tipos de composto) e 8 repetições. Cada unidade experimental constou de uma caixa com 12–12,5 kg de composto fresco úmido. Os dados foram submetidos à análise de variância e as médias foram comparadas pelo teste de Tukey. De acordo com os resultados obtidos verificou-se que a produção de cogumelos foi influenciada pela linhagem e/ou pelo tipo de composto. Também verificou-se que o teor de proteína bruta, cinzas e fibra bruta de basidiomas variou com a linhagem de *A. bisporus* e com o tipo de palha utilizada na formulação do composto.

**Palavras-chave:** *Agaricus bisporus*, linhagens, palha, composição nutricional, produtividade.

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